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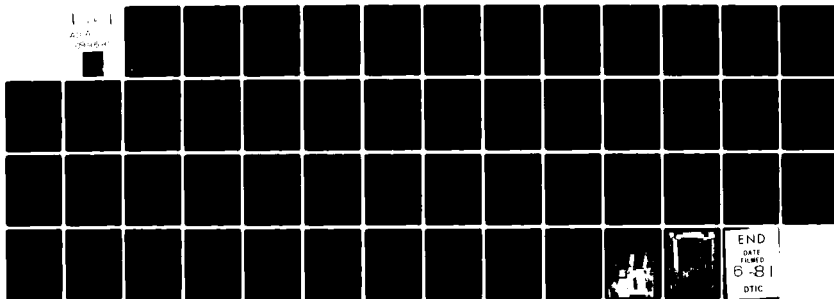
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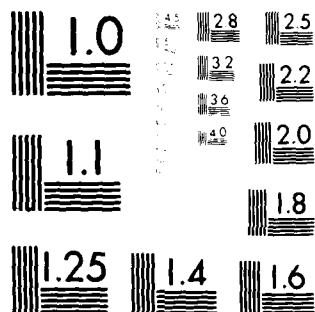
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On Project

Physiological Adaptations of Arctic Mammals.

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1a. Introduction

This program was in two phases: four experiments were totally new; several which were long-range experiments were brought to completion; and one important one which we intended to begin will now be terminated.

Experiments which were totally new were: a) The Mechanism of Increased Tolerance of Rodents Exposed to -40° (for details see Table of Contents, #9 and Part F in Expenditures); b) The Effect of Norepinephrine on Muscle O_2 Consumption (see Table of Contents #s 10 and 11, and Part E in Expenditures); c) The Day-Night Core Temperatures of Wolverines and Weasels (see Table of Contents #12 and Part B in the Expenditures); d) Studies on the Pineal Gland of Arctic Animals (see Table of Contents #7 and Part J, Item 7, and Part C, the item for \$1,090 in the Expenditures).

Those experiments which were carried out over several years and are now brought to completion or will be finished in 1976: a) Cold Acclimation in Lemmings and Voles (see Table of Contents #s 1, 2, 3, and 4, and Item G in Expenditures); b) Studies on Hibernation Factor Transferred from Marmots to Test Animals (see Table of Contents #5 and Item I in Expenditures); c) Fat Analysis of Marine Mammals (seals and polar bears) Including Determinations of Serum Lipids and Ketone Bodies in Urine of Polar Bears (see Table of Contents #6 and Parts D and H in the Expenditures); d) Leadership in Wolves with Telemetry of Cardiovascular Indicators and Observations on Reproductive Dominance (see Part C in Expenditures); e) Measurement of Energy Metabolism and Daily Energy Budget by Heart Rate as an Indicator (see Table of Contents #s 8, 13, and Parts B and C in Expenditures).

The experiment which we intended to begin but will now be terminated is as follows: the experiments asked whether seals have a physiological tidal rhythm which is different from a 24-hour rhythm, and also whether seals normally and spontaneously demonstrate diving bradycardia to the extent that this is portrayed in the literature. The reason for the last question is that many studies on the slowing of the heart beat in seals have been done artificially in the laboratory, often by forcing the seal underwater. This was a radio telemetry program. A permit was obtained to maintain four Ring seals at the Naval Arctic Research Laboratory. For details for this part of the experiment, see Part A in Expenditures. A second part of the experiment, however, involved taking samples of blubber over four seasons to determine the changes in composition. Not only is blubber very depleted but it probably becomes watery. Some data on this part of the experiment was obtained by collecting blubber from seals shot by Eskimos on the Arctic ice pack. This experiment was extremely well done by Keith Magoon who took dry ice and nitrogen out on the ice so that he could collect and preserve extremely fresh blubber samples (see Part A and Part J, Item 11 of Expenditures). This project was not carried out and

Introduction (continued)

will be terminated because of the difficulties of maintaining the seals. Not only did the Utilidor, which was to deliver water to the tanks, break down, but also the original plan of delivering water by Utilidor to the Animal House has now been abandoned.

In the case of each topic discussed above, explanations of the applications and the usefulness of each experiment to the Office of Naval Research are included in each numbered item found in the body of this report, and listed in the Table of Contents.

Title: Cold Acclimation in Arctic Lemmings*

Authors: J.J. Berberich and G.E. Folk, Jr.

Journal: Comparative Physiology and Biochemistry

Status: Accepted; in press, pages four to six.

Coordination with Program: For the last four years, we have been studying cold acclimation in lemmings, from many approaches and aspects. This manuscript includes about one-fifth of our data on this topic, obtained in the last two years. Of course, my motivation for studying cold acclimation in lemmings was a question as to whether arctic animals show cold acclimation or whether they are in this condition or state all year round.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

ABSTRACT

1. The lemmings, arctic rodents, show cold acclimation as measured by a number of variables: food and water consumption, liver temperature, and organ weights.
2. Cold acclimation of lemmings is apparently more rapid than that of temperate mammals.
3. The rapid cold acclimation in lemmings supports the hypothesis that compensation of a number of physiological variables is essentially complete within the first week of cold exposure as opposed to requiring months.

INTRODUCTION

Initial physiological responses to cold and cold acclimation have been studied extensively in temperate mammals, notably the white rat (Hérault, 1961) and hamster. Arctic mammals, however, may serve as better models of cold acclimation since they naturally experience lower temperatures. The opportunity was afforded to investigate at the Naval Arctic Research Laboratory, Barrow, Alaska the cold acclimation of two species of arctic rodents: the brown lemming, Lemmus trimucronatus, and the varying lemming, Dicrostonyx groenlandicus. These two species are the most northerly distributed North American small mammals and are unique in being endemic to the arctic as genera (Schwartz, 1962). This report presents data of some basic physiological indices of cold acclimation in the lemmings which may be compared readily to those of temperate mammals.

Title: Tolerance of Arctic Lemmings to Hypothermia and Dehydration*

Authors: J.J. Berberich and G.E. Folk, Jr.

Journal: Cryobiology

Status: Submitted; six pages of manuscript.

Coordination with Program: The introduction of this paper relates these tests of hypothermia and dehydration to our overall program on Arctic mammals. I have personally seen lemmings fall in ice water during temperatures near freezing. This must happen fairly frequently, and the question is whether these animals can survive after being challenged by this severe environment. In addition to this, we are especially interested in that period when lemmings must lick ice to obtain water. One would think this is a handicap to their temperature regulation. This explains the experiment written up in the next two pages.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

TOLERANCE OF ARCTIC LEMMINGS TO HYPOTHERMIA AND DEHYDRATION

Measurement of the time to death due to exposure to an environmental stressor has classically been used to demonstrate the ability of mammals to withstand the stressor, usually without including measurements to explain how tolerance is acquired (Hart, Comparative physiology of thermoregulation, vol. II, p. 105, 1971). This approach is useful to compare the relative tolerances of different species and to evaluate the effects of acclimation.

The brown lemming, Lemmus trimucronatus (= sibiricus), and the varying lemming, Dicrostonyx groenlandicus, are the most northerly distributed small mammals in North America. The environment they inhabit is characterized by low temperature year-round, an abundance of free water on the tundra during the summer, and snow in the winter. There are periods before snowfall when lemmings must lick ice to obtain water. Thus, one might meaningfully characterize their physiological ecology by a hypothermia test and a dehydration test.

Animals were bred and maintained at the Naval Arctic Research Laboratory animal colony (Barrow, Alaska) at thermoneutral temperature (18°C) on a summer photoperiod (22L:2D). Some animals were cold acclimated by exposure to 3°C for 17-26 days on a summer photoperiod.

Hypothermia test. Animals were exposed to -40°C in a cold box (Forma Scientific) inside small cages, large enough to turn

The lack of dehydration tolerance of the lemmings is the most noteworthy feature of that experiment. These results are striking because of their similarity to the tundra vole, Microtus oeconomus. German (Zool. Zhur., in Russian, 40:914-924, 1961) found that the tundra vole tolerated dehydration with dry food for 2-3 days with a body weight loss of 25%. Lack of dehydration tolerance may be a common feature of rodents inhabiting the tundra. Note that German found other species which could tolerate nearly 40-50% weight loss during dehydration. The organ water content of the dehydrated lemmings in this study was compared to control values. Significant water loss was observed for lung, spleen, and muscle (gastrocnemius) for both species, but was most remarkable for the liver: 25% for the brown lemming and 14% for the varying lemming.

In summary, the physiological responses of the lemmings are indicative of the environment they inhabit. They show superior tolerance to severe cold and poor tolerance to the lack of water.

This study was supported by The Arctic Institute of North America under contract with The Office of Naval Research, and by the National Science Foundation. Many of the varying lemmings used in this research were graciously provided by Professor Edwin M. Banks of the University of Illinois.

J. J. Berberich and G. E. Folk, Jr., Department of Physiology and Biophysics, The University of Iowa, Iowa City, Iowa 52242.

Title: Renal Function in Hypothermic Arctic Lemmings*

Authors: J.J. Berberich and G.E. Folk, Jr.

Journal: Journal of Mammalogy

Status: Submitted; four pages of manuscript.

Coordination with Program: The most striking indicator of cold acclimation in lemmings turned out to be a massive diuresis which at times is represented by an amount of urine equal to the weight of the animal. In the experiment described in this paper, it was found necessary to expose brown lemmings to -40°F . The reason for this was that it has not been understood in the past whether acclimation at 5° or at least some observations which we call acclimation, actually prepares the animal for exposure to an extremely cold environment. In other words, we may call a group acclimated, but they might not tolerate -40° any better than the control animals. While doing this experiment, some very surprising observations on kidney function were noted. They are presented as abstracted on the next few pages.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

RENAL FUNCTION IN HYPOTHERMIC ARCTIC LEMMINGS

Progressive depression of renal function during hypothermia generally is presupposed. Data gathered incidentally as part of a detailed study of arctic lemmings (Berberich, Ph.D. Thesis, University of Iowa, 1975; Berberich and Folk, Comp. Biochem. Physiol., in press) suggest that renal function in hypothermic arctic lemmings ought to be investigated in detail.

Unacclimated and cold-acclimated (3°C, 2 weeks) brown lemmings (Lemmus trimucronatus sibiricus) and cold-acclimated varying lemmings (Dicrostonyx groenlandicus) were exposed in small stainless steel cages to -40°C ambient temperature in a cold box. Most animals were kept at -40°C for 55 minutes of each hour until they lost the ability to right themselves when placed on their backs. Mean exposure times were 2.6 hours for unacclimated brown lemmings, 15.6 hours for acclimated brown lemmings and 20.7 hours for acclimated varying lemmings. The urine voided by these severely cold exposed lemmings was collected at either half hour or hour intervals on clean plastic (Parafilm). This frozen urine then was placed under mineral oil for subsequent analysis by freezing point depression for osmolality and by flame photometry for sodium and potassium concentration.

Data for the first 30 minute period of severe cold exposure is summarized in Table 1. All the urine concentrations are

significantly decreased compared to values of control (18°C) and cold (3°C) exposed animals (Berberich and Folk, Comp. Biochem. Physiol., submitted). However, sodium to potassium ratios and sodium plus potassium to osmolality ratios are not different. At any rate, a depression of renal concentrating ability is demonstrated with initial severe cold exposure.

A paradox, however, is found with prolonged severe cold exposure. Although core temperatures were not continuously measured, at the time of removal from the cold box, liver temperatures were between 15 and 20°C. In spite of the fact that the animals had to be experiencing progressive hypothermia, concentrating ability did not change with prolonged severe cold exposure (Table 2). Data are presented as means of individual regression equations to account for the differential lengths of cold exposure. With the exception of the slope of potassium concentration of unacclimated brown lemmings (which increased), slopes of ion concentrations were not different from zero. No change in sodium and potassium concentrating ability developed with prolonged severe cold exposure and progressive hypothermia.

Although these data are limited, lemmings apparently show renal physiological adjustments to hypothermia. This possibility ought to be investigated in detail.

This study was supported by The Arctic Institute of North America under contract with the Office of Naval Research, contract # N00014-72-A-0375-0002 (subcontract ONR-453), and by The National Science Foundation.

Title: Cold Acclimation in the Tundra Vole (Microtus oeconomus)

Authors: Peter J. Ringens, G. Edgar Folk, and Joel J. Berberich

Journal: The European Journal of Physiological Mammalogy (Acta Theriologica)

Status: Accepted for publication; nineteen pages of manuscript.

Coordination with Program: This experiment turned out to be a very important spin-off. When I left for Barrow in 1974, the Dutch government had sent a medical student to spend a few months in my laboratory. I had no recourse except to bring this medical student to Point Barrow. He became completely dedicated to Arctic science, and his goal in life is to get his M.D. degree and to come to Point Barrow to practice medicine there, meanwhile continuing experiments at NARL. He turned out to be a work-horse. His job was to clean all glassware for ultra-chemistry determinations. He also had to take part in collecting urine from lemmings and making measurements at four different times of the 24-hour period. Although the day was already extremely full, another experiment developed. Our question concerned whether arctic animals show cold acclimation. It was conceivable that arctic mammals, especially small rodents which live in cool, wet runways, might be in a state of cold acclimation the entire year around. While we worked on the lemmings, the animal colony developed by breeding, a surplus of the tundra vole. Here was our opportunity to check a second arctic animal to see if it would show cold acclimation at all. I asked Ringens if he would take on extra work as his own problem to do with me. He eagerly accepted and here is his manuscript.

Credit: Same as the previous publications.

Cold Acclimation in the Tundra Vole (*Microtus oeconomus*).

Peter J. Ringens^{*}, G. Edgar Folk^o, Joel J. Berberich^o.

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Physiology and Biophysics, Iowa City, Iowa.

Abstract.

Two groups of tundra voles (*Microtus oeconomus*) were exposed both to control temperature (17° C) and to cold (4° C).

Some physiological parameters involved in adaptation to the cold, such as cold diuresis, food consumption, weight loss, urinary pH, Na and K were studied.

Cold diuresis was found, stabilizing on a certain level after a few days; the same was found for the food consumption.

The animals lost weight during the first two weeks of cold exposure, and then began to recover until they reached their original weight again.

The pH of the urine dropped during the first days in the cold, but recovered after approximately 15 days. A circadian pattern for the pH was recorded, showing an increase during the 24-hour cycle, with its maximum between 5:00 AM and 9:00 AM.

Interpretation of the sodium and potassium excretion in the urine is difficult as long as there are no specific data on "intake" and "balance" available.

However, the data can be interpreted in relationship with the pH data.

Title: Induction of Summer Hibernation in the 13-Lined Ground Squirrel Shown by Comparative Serum Transfusions from Arctic Mammals*

Authors: Wilma A. Spurrier, G. Edgar Folk, and Albert R. Dawe

Journal: Cryobiology

Status: Accepted; sixteen pages of manuscript.

Coordination with Program: For the last five years, we have had a continuing program of extracting hibernation blood from arctic rodents, bringing it to The University of Iowa, processing it, and testing it either in this laboratory or in the laboratory of Dr. Albert Dawe. I presented one control experiment which has not been done by Dr. Dawe, namely, that the hibernation factor may be there because the animal is cold, not because it is hibernating. This would mean that a hibernator under hypothermia or even a non-hibernator under hypothermia, might have blood which acted as a hibernation substance or "trigger". This paper settles that question, along with proof that relatives of the woodchuck (i.e. arctic marmots) also can produce hibernation factor.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

Previous reports from this laboratory summarized in (2) have demonstrated that there is a "trigger" for induction of hibernation present in the blood of animals in the state of hibernation. This trigger substance is of a molecular weight smaller than 10,000 Daltons, and has been found in whole blood, serum, or dialysate of serum. The trigger is extractable when the animal is hibernating, but is absent from blood drawn from animals arousing from hibernation, and absent from summer active hibernators. The donors and recipients in previous experiments were the 13-lined ground squirrel (*Citellus tridecemlineatus*) and the woodchuck (*Marmota monax*). Since these two species are strict seasonal hibernators any departure from the events of the usual circannual rhythm is significant. By administering hibernation bloods and their derivatives in a season when the animals would not normally hibernate we were able to utilize summer hibernation at a behavioral "end point" for studying hibernal-induction effects. As reported elsewhere (3), the serum of spring and summer active hibernators contains a nondialyzable larger molecular weight (greater than 10,000) substance in high concentration, which can be shown to negate effect of the hibernation trigger and is therefore called "anti-trigger". If the anti-trigger substance is injected into active hibernators in mid-summer it does not induce summer hibernation, but it impedes the occurrence of hibernation in the fall by as much as a month or two. This experimental evidence suggests that there is a balance between trigger and anti-trigger, which determines whether an animal remains active, or hibernates.

An interesting aspect of the hibernation trigger is that it is not species specific. Bloods and blood derivatives of 13-lined ground squirrels and of woodchucks have been cross-transfused in small amounts without adverse reactions. Hibernation can be induced between these two species. Therefore, one of the objec-

SUMMARY

A "trigger" substance was again demonstrated to be present in sera of hibernating animals. The hibernating 13-lined ground squirrel, hibernating woodchuck, hibernating Arctic ground squirrel and hibernating Arctic marmot, were all capable of inducing the 13-lined ground squirrel to hibernate in the summer, a season when that species would normally be active. The hibernation trigger is thus not species specific. It is effective whether drawn from these two Arctic species of hibernators, or drawn from these two species of hibernators from the mid-western states. The normothermic Arctic marmot appears to have an "anti-trigger" substance in its serum in the summer, which impedes fall hibernation in the transfused 13-lined ground squirrel. This is similar to the anti-trigger observed in the summer serum of active 13-lined ground squirrels and active woodchucks. With respect to hypothermia, it was induced in Arctic marmots and in Arctic foxes at Point Barrow, Alaska in summer. Though in such cases body temperatures fell significantly (as in hibernation) no trigger was recovered from their hypothermic sera that could be shown to be capable of inducing summer hibernation in the ground squirrel. Neither was anti-trigger found in the serum of hypothermic experimentals. These latter experiments thus suggest that the release of trigger into the blood during hibernation is dependent on a mechanism more complex than simply lowering body temperature.

Title: Characterization of Polar Bear Serum Lipoproteins*

Author: Terry L. Kaduce

Journal: This paper was a Masters Thesis in the Department of Biochemistry in this Medical School.

Status: In Thesis form; seventy-eight pages.

Coordination with Program: For a number of years, by cooperation between several different laboratories, we have surveyed the status in the serum of arctic animals of lipids. By cooperation with our Department of Biochemistry, with these samples of polar bear serum, which I took from the fasted animal one week apart, with triplicate samples, we have been able to make some thirty determinations of the components of serum lipids in this animal, with an emphasis upon the lipoproteins. Shortly I will write a paper with Mr. Kaduce for the Journal of Comparative Biochemistry and Physiology on this material.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

SUMMARY

The serum lipoproteins of polar bears living in their natural Arctic environment were characterized and compared to human serum lipoproteins. The fatty acid composition of the lipids associated with the bear lipoproteins were analyzed and found to contain less linoleic and more eicosapentanoic acid than normally found in man or most other mammals. Polar bear serum triglyceride and cholesterol concentrations were found to be similar to those in human plasma, but phospholipid levels were found to be elevated. Polar bear serum contained only very small amounts of VLDL. The LDL from these bears were comparable to human LDL in cholesterol and phospholipid content, and their apoprotein composition was similar to human LDL both by SDS-polyacrylamide gel electrophoresis and by amino acid composition. However, the polar bear LDL contained a much higher triglyceride content than either human LDL or those of other species. Polar bear HDL lipids are present in higher concentrations but in the same relative ratios as in human HDL. The apoproteins of the polar bear HDL had similar SDS-polyacrylamide gel electrophoretic properties as compared with human apo HDL.

Title: Photoperiodism and the Physiology of the Pineal Gland*

Author: G.E. Folk, Jr.

Journal: International Journal of Biometeorology, 1975, vol. 19,
number 4, pages 217-218.

Status: The enclosed is the Preface to an entire Symposium,
published in entirety; Vol. 19, pages 217-310.

Coordination with Program: In 1974, we began making measurements on the pineal glands of lemmings at NARL. This seemed appropriate since this gland represents an organ which is reputed to be entirely associated with changes in daylight and darkness; of course in the Arctic with 82 days of continuous light and continuous darkness, it appeared that there might be some unusual findings concerning the pineal gland. When I was asked to organize a symposium concerned with photoperiodism for the 7th International Biometeorological Congress, I decided that to emphasize the pineal gland would be most appropriate since its relationship to extreme light cycles has received little attention. This symposium will serve as a synthesis and a review of the literature for our continuing work on the pineal gland at NARL.

Credit: * The organization of this symposium and the presentation of two of the papers was supported by a grant from The Arctic Institute of North America with the approval and financial support of the Office of Naval Research.

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SYMPOSIUM

on

PHOTOPERIODISM AND THE PINEAL GLAND

Held during the 7th International Biometeorological Congress,
17-23 August 1975, College Park, Maryland, USA

Chairman: G.E. Folk Jr.

Reports edited by: D.C. Dawson

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Photoperiodism and the Physiology of the Pineal Gland

by

G.E. Folk Jr.*

INTRODUCTION

The symposium "Photoperiodism and the Pineal Gland" was organized in the perspective of trends in Biometeorology over the last nine years. There are several approaches to environmental relationships; one may select a biological phenomenon, i.e. reproduction, and inquire as to its relationship with the physical environment. Conversely, we may select some aspect of the total physical environment and study its impact upon animals and plants. For many years the International Society of Biometeorology, in its journal and congresses, has emphasized the second approach; the titles of journal articles and congress sessions have included the effects of temperature, low pressure, humidity, and other more subtle environmental factors upon animals and plants. In addition the phenomena referred to collectively as "Biological Rhythms" have captured the attention of members of this society. Interestingly, however, the environment of light *per se* has received little attention. It was the function of this symposium to treat the topic of Photoperiodism as a separate entity, and in addition, to probe more deeply into one emerging topic in photoperiodism, the physiological role of the pineal gland. There was an attempt to include a spectrum of photoperiodic phenomena, i.e. seasonal cycles as well as daily rhythms. The topic "The Pineal and Daily Rhythms" had been suitably reviewed in two recent symposia which evolved spontaneously and simultaneously with our own, one at the American Society of Zoologists' meeting (1973), and one at the Federation Meeting (FASEB, 1974).

The perspective on pineal function has been enlarged by the demonstration of photoperiodic effects which in the past were not suspected of involving this gland. A product of the pineal gland, melatonin, has been shown to have a relationship to hibernation. Palmer and Riedesel (1975), have demonstrated that animals in hibernation go in more rapidly and have longer bouts of hibernation under the influence of melatonin. In addition, light is now known to influence reactions to cold so that we can say "it is possible to have cold acclimatization without cold" (Lynch and Folk, 1971). These new aspects must be integrated with the known effects of the pineal gland upon reproduction. Results from the laboratory of R.J. Reiter illustrate dramatically the role of the pineal gland in the seasonal gonadal adjustments of the hamster. Exposure of hamsters to "short days" (2L:22D) results in profound testicular regression from approximately 3,000 mg to about 500 mg. This short day response is abolished by pinealectomy. In fact, the gonads of blinded hamsters remain fully functional if the animals are pinealectomized (Reiter, 1974).

It was decided in this symposium to organize our ideas in part by reasoning from the physical environment to biological principle, that is to ask, what the relationship of various lighting regimens, in particular continuous light and con-

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tinuous darkness, would be to rhythmic phenomena and pineal function. It was hoped that this might provide some insight into possible mechanisms of adaptation to extreme variations in natural photoperiods, such as those experienced by hibernating animals and those which form part of the seasonal arctic cycle. Dr. Charles Ralph consented to consider the relationship between latitude and pineal gland physiology. An Arctic physiologist was invited to review the reproduction of arctic animals for the following reasons: those circum-polar mammals which migrated over the land bridge from Siberia might very well be expected to have totally different photoperiodic responses compared to the group of animals which at a much older time evolved near the equator and spread north toward Canada. The living circum-polar (Russia, Siberia, Alaska) mammals, each a single species, include wolverine, lynx, wolf, reindeer, and varying lemming. Because these animals must tolerate extremes in photoperiod (in the same fashion as Reiter's hamsters), we were pleased that the scientific director of the Naval Arctic Research Laboratory at Point Barrow, Alaska, was able to contribute his topic: "Continuous Light and Physiology of Arctic Birds and Mammals".

In summary, in this symposium volume one may expect to find a broad approach to, and a wide-ranging analysis of, the effect of the "photic" environment upon animals, and in particular on the physiology of the pineal gland. The organization of this symposium and the presentation of two of the papers was supported by a grant from The Arctic Institute of North America with the approval and financial support of the Office of Naval Research under contract number N00014-75-C-0635 (Subcontract ONR 455).

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ABSTRACT
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Title: Simultaneous Measurements of Heart Rate and Oxygen
Consumption in Black-Tailed Prairie Dogs (Cynomys
ludovicianus) *

Authors: Gordon F. Lund and G. Edgar Folk, Jr.

Journal: Comparative Biochemistry and Physiology

Status: Accepted; twenty-one pages of manuscript.

Coordination with Program: This experiment has a very interesting history associated with Arctic science. This student wished to do his Ph.D. work in my laboratory and at that time we had four arctic ground squirrels. Because it is of importance to the Navy to know the energy budget of those animals living on Petroleum Reserve 4, it seemed reasonable to me to attempt to estimate on arctic animals their oxygen consumption by means of heart rate radio telemetry. Others have done this on other species. We began work successfully with the four arctic ground squirrels. Before submitting them to operations, I attempted to breed them so that we would have a continuing supply; this was unsuccessful and we went ahead and implanted these animals. For two reasons it was necessary to stop work on these animals: firstly, NARL tried unsuccessfully for a spring, summer, and fall to obtain arctic ground squirrels for us. Although it seems strange, they were unable to do this. Also, we could not build a suitable outdoor free-run area in which we could release these animals.

We therefore turned to a nearby colony of prairie dogs, since they not only are the same weight as the arctic ground squirrels, but they behave the same way, and we felt that they could serve as a model of the arctic animal. It is of interest that they have now been shown to hibernate in the same way that arctic ground squirrels do. A Ph.D. thesis was completed which we believe can show some carryover to measurements which can now be made of the energy budget of large rodents in Petroleum Reserve 4. This manuscript represents approximately one quarter of the work that was accomplished. The rest is in manuscript form as a monograph and we are searching for a publisher.

Credit: Same as other publications referred to previously.

ABSTRACT

1. Studies were conducted to explore the potential for using telemetry of heart rates as an index of rates of energy expenditures in free-ranging black-tailed prairie dogs (Cynomys ludovicianus).

2. For periods of one hour with activities varying spontaneously or by exciting the animal, heart rate and oxygen consumption were highly correlated with coefficients averaging above 0.9.

3. The relationships appeared stable for one to a few days but differed over weeks or longer.

4. Relative comparisons of costs for behaviors over circadian cycles and a few days would seem feasible by the heart rate index method in prairie dogs.

Title: Oxygen Consumption and Body Temperature Measurements of
Lemmings Maintained at -40° *

Authors: G. Edgar Folk, Jr. and Olivier Héroux

Status: Manuscript in partial and preliminary form.

Coordination with Program: The essence of this experiment is that it was found at the National Research Council of Canada that there was a wide variation in standardized white rats as far as their resistance to -40° is concerned. They wished to repeat this experiment on an arctic mammal in hopes of analyzing the short-term resistant animals and the long-term resistant animals to discover what mechanism permits the extra resistance. They asked to collaborate with our work on lemmings and agreed to send Dr. Olivier Héroux with some unique equipment which can measure oxygen consumption of animals maintained at -40°. They were going to pack up this elaborate equipment and set it up again at NARL for the months of July and August. We proceeded with this plan. Determinations such as the free fatty acids and the glycogen, and corticosteroids, are still being made from many samples taken from a large number of lemmings. On the next few pages, the preliminary planning is presented, followed by three samples of the many pages of data which were obtained. The manuscript will be written when Dr. Folk joins Dr. Héroux in late spring to do the final interpretation of the data.

Credit: * Supported by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

In the Words of Dr. Folk:

The background of this request is based upon our work at Point Barrow each summer and winter since 1960. The essential theme of our endeavors has been the elucidation of adaptations of Arctic mammals to extreme cold. Studies on the biological clocks of these animals and especially acclimatization to cold have been in the forefront of our experiments.

Lately we have been concentrating upon the details of cold acclimatization in lemmings, since we wished to compare them with temperate zone mammals, specifically white rats and human subjects. We believe that there might be some special mechanisms in those species which have survived the Arctic climate for many thousands of years.

We have some evidence to show that the brown lemming, available at Point Barrow, expresses a totally new form of cold acclimatization. Specifically, we found that the kidney responds in a curious fashion so that the cold-exposed animal often produces its own weight in urine per day. However, we found quite a bit of variation between individual animals in their tolerance of extreme cold. In surveying the literature, we found that the authority in this area is your Dr. Olivier Héroux. I had read his papers concerned with cold resistance under severe Arctic conditions for many years. During the process of his authoritative work in this area, he has perfected very workable techniques for accurate measurement of oxygen consumption of animals maintained at -40° C. Because we are inexperienced in this area, we are rather eager to combine resources with him this summer during some period when he might be permitted to work in the laboratory at Point Barrow. Hopefully this might be part of, or all of, the month of July. The details of the design of the experiment (which I think will make a large fundamental and theoretical contribution to cold weather physiology) are included in the enclosed memorandum. The major question in this letter is whether he may join us at the Naval Arctic Research Laboratory.

I think I can safely say that without his expertise we will not be able to attack the question as to why only certain individual animals can maintain their body temperature under the challenge of cold. In other words, his presence and his contribution of part of the equipment would be the decisive point as to whether this research can be begun.

In the Words of Dr. Olivier Héroux:

As I told you in Yellowknife and as you will see in the copy of the manuscript that I am sending you, we found very little variation in the level of O_2 uptake reached by individual $6^\circ C$ acclimated rats exposed to $-40^\circ C$, but the length of time each rat could maintain that level varied greatly from one individual to the other. Moreover, we found, by continuously monitoring colonic temperature, telemetrically, that in every case body temperature dropped rapidly to approximately $32.9^\circ C$ where it plateaued to fall rapidly again when metabolism failed.

These observations raised the interesting question as to why certain individual animals would be able to maintain a new equilibrium at $32.9^\circ C$ for only 10 min or so, while others would be able to do so for 50 min and some others for 150 min. Is it due, for example, to the exhaustion of substrates (FFA or glycogen) or the exhaustion of hormones (corticoids) and if so, why would that happen sooner in one individual than in another one? We found in adrenalectomized rats that without adrenals, a cold acclimated rat could still reach essentially the same level of O_2 uptake as the intact animals, but he could not maintain it for any length of time so that its resistance time was greatly reduced.

If you are wondering if small mammals from the Arctic zone, namely the lemming, would under similar circumstances ($-40^\circ C$), 1) show similar individual variability in their cold resistance, 2) maintain a new equilibrium at a similar body temperature ($32.9^\circ C$), 3) be similarly affected by the removal of their adrenal, I would gladly collaborate with you in that study, especially since you seem to have all the facilities required to do these experiments.

I would welcome the opportunity 1) to repeat our study on an arctic mammal, 2) to extend our study by determining the level of FFA in blood and glycogen in liver and muscle at the failure point to see if there is any correlation between these levels and the resistance time, 3) to verify if the damaging effect of adrenalectomy in the capability of maintaining a high metabolic rate is due to the absence of corticoid secretion or catecholamine secretion which could be verified by removing only the medulla in certain animals and the whole adrenals in others.

The experiment would essentially consist in cold acclimating lemmings to 5°C for 4-5 weeks and then exposing them to -40°C in a presto cooker immersed in an antifreeze bath and connected to a closed-circuit system for measuring O₂ uptake. I could bring with me the closed-circuit respirometer that we have used; it is very efficient at very low temperatures and moreover, I am familiar with it. It has been described in J. Appl. Physiol., 28: 712-713, 1970. Each animal would have to be fitted, a few days before with a transmitter for monitoring body temperature. Judging from the B.T. curve, it will be possible to know when the metabolism has failed, at that time, the animal will be sacrificed and blood as well as liver and muscles will be collected and frozen in liquid nitrogen to be analyzed later on.

Date (Run)	Animal Number	Treatment	Caudal Fat Pad		Brown Fat Pad		Weight of Stomach Contents (Grams)
			1	2	1	2	
11/10/75	Pilot B	Non Cold Acclimated at -40 (T)	.060	not taken	.055	not taken	.068
11/11/75	Pilot C	Non Cold Acclimated at -40 (T)	.090	not taken	.065	not taken	.127
11/11/75	# 30 ^{SNIP}	Cold Acclimated no -40 (T)	.069	.060	.033	.041	.061
11/12/75	# 40 ^{SNIP}	Cold Acclimated no -40 (T)	.037	.061	.143	.089	.141
11/13/75	# 34	Cold Acclimated at -40 (T)	.048	.069	.073	.112	.434 (huge solid air ball filling stomach)
11/14/75	# 36 ^{SNIP}	Cold Acclimated no -40 (T)	.066	.078	.112	.130	.045
11/14/75	# 26	Cold Acclimated at -40 (T)	.088	.092	.070	.054	.108
11/14/75	# 16	Cold Acclimated at -40 (NT)	.079	.029	.077	.092	.075
11/15/75	# 37 ^{SNIP}	Cold Acclimated no -40 (T)	None	None	.059	.072	.039
11/15/75	# 33	Cold Acclimated (T) DIED	—	—	—	—	— Died before -40 run
11/15/75	# 38	Cold Acclimated at -40 (NT) DIED	—	—	—	—	— Died in chamber (drowned)
11/16/75	# 13	Cold Acclimated at -40 (NT)	.093	.062	.042	.084	.239
11/17/75	# 28	Cold Acclimated at -40 (T)	None	None	.045	.051	.867
11/18/75	# 20 ^{SNIP}	Cold Acclimated no -40 (NT)	.198	.140	.141	.127	.253
11/19/75	# 21 ^{SNIP}	Cold Acclimated no -40 (NT)	.052	.046	.065	.084	.316
11/19/75	# 33	Cold Acclimated at -40 (NT)	.123	.102	.103	.109	.087
11/19/75	# 9 ^{SNIP}	Cold Acclimated no -40 (NT)	.099	.152	.121	.153	.2062
11/21/75	# 1	Cold Acclimated at -40 (NT)	.528	.321	.096	.077	.063
11/22/75	# 10 ^{SNIP}	Cold Acclimated no -40 (NT)	.482	.368	.322	.315	1.056
11/22/75	# 3	Cold Acclimated at -40 (NT)	.209	.116	.102	.064	.188
11/23/75	# 11 ^{SNIP}	Cold Acclimated no -40 (NT)	.166	.247	.054	.032	.523
11/24/75	# 22 ^{SNIP}	Cold Acclimated no -40 (NT)	.329	.305	.257	.274	.158
11/25/75	# 23 ^{SNIP}	Cold Acclimated no -40 (NT)	.400	.193	.144	.179	.746
11/25/75	# 18	Cold Acclimated at -40 (NT)	.146	.121	.131	.123	.097
11/26/75	# 24 ^{SNIP}	Cold Acclimated no -40 (NT)	.092	.046	.045	.074	.136
11/26/75	# 39	Cold Acclimated at -40 (NT)	.122	.094	.169	.166	.820

Table 2: Oxygen Consumption of Lemmings ($T_A - 40^\circ$)

WARM: 19°C .

COLD: 5°C

WITH TRANSMITTER		WITHOUT TRANSMITTER				WITH TRANSMITTER				WITHOUT TRANSMITTER			
BWT	O^2	RESISTANCE	Nb	O^2	RESISTANCE	Nb	P.Wt.	O^2	RESISTANCE	Nb	P.Wt.	O^2	RESISTANCE
(g)	(ml/kg) (ml/kg)	(min)		(ml/kg) (ml/kg)	(min)		(g)	(ml/kg) (ml/kg)	(min)		(g)	(ml/kg) (ml/kg)	(min)
57.8	3.1	35	?	3.5	?			3.5	?			3.5	?
47.6	4.8	55	1 ♀	5.6	55	1 ♀	57.7	10.4	10.5	15	34	5.2	7.0
44.3	4.2	25	3 ♂	6.3	25	3 ♂	66.3	9.0	8.1	30 (55)	26	4.8	6.3
			5 ♂	6.50	50	5 ♂	65.0	9.0	8.2	50	28	4.1	7.2
			<u>Specimens</u>										
			♀	33.4	70	12.6				45 > 70			
			♀	31.6	47	9.5				30			

Table 3: Measurements on Fresh Wild-Caught Male Lemmings;
Control Values to Compare with Captive Animal Values.

animal #	Hematocrit	plasma Na	plasma K
1	-	132.5	5.8
2	45.5;	124.7	10.7
3	45.7; 46.0	-	-
4	49.5; 50.0	126.9	9.3
5	46.0;	124.4	10.2
6	45.0; 44.7	137.7	8.4
7	44.5;	134.9	7.4
8	46.0; 47.5	129.6	11.3
9	-	124.8	5.2
10	47.8;	132.2	9.9
11	45.0;	124.9	10.6
average:			
	46.2 \pm 0.6	129.3 \pm 1.6	8.9 \pm 0.7

Title: Effect of Norepinephrine on Muscle O₂ Consumption of Cold Acclimated Rats *

Authors: Barbara Grubb and G.E. Folk, Jr.

Journal: Federation Proceedings, Vol. 35, page 559 (1976)

Status: Published

Coordination with Program: This experiment was part of our studies of cold acclimatization in the lemming. We believe there has been an over-emphasis on the contribution of brown fat to the thermogenesis of the cold acclimated animal. We have perfected new and elaborate equipment which can perfuse the hind limb of the rodent. It was necessary to do that first on a larger animal-- the white rat. We have, however, tried this technique on four arctic lemmings with complete success. The new part of our program will consist of cold acclimatizing lemmings and repeating work which has been done on the rats, in the same way.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

EFFECT OF NOREPINEPHRINE ON MUSCLE O_2 CONSUMPTION OF COLD ACCLIMATED RATS. Barbara Grubb* and ²G. E. Folk, Jr. Univ. of Iowa, Iowa City, Ia. 52242

The administration of norepinephrine (NE) causes a greater increase in oxygen consumption ($\dot{V}O_2$) in rodents acclimated to cold than in control animals. Much of the increased oxygen consumption has been attributed to skeletal muscle. To determine if muscle from cold acclimated rats has an increased sensitivity to NE, the rate of oxygen consumption by the perfused hindlimb (constant-flow) of cold acclimated and control rats was investigated. The oxygen content of the perfusate was determined by oxygen electrode. Infusion of NE (.0001-.1 $\mu\text{g/g/min}$) for five minutes resulted in an immediate and similar rise in $\dot{V}O_2$ in both cold acclimated and control limbs. Infusion of NE (.01 $\mu\text{g/g/min}$) for 30 min resulted as above in an immediate increase in $\dot{V}O_2$ by both groups of muscles; the $\dot{V}O_2$ of the cold acclimated muscle remained elevated throughout the 30 min infusion period, while the $\dot{V}O_2$ of the control muscle declined continually, not being significantly above the unstimulated level at the end of the 30 min infusion period. These results suggest that cold acclimation does not change the magnitude of the initial NE stimulated increase in muscle oxygen consumption, but a sustained elevated $\dot{V}O_2$ was found only in the muscle from the cold acclimated rat. The technique is satisfactory for a study in progress using small (30-80g) arctic lemmings. (Supported by ONR for AINA, and NIH Postdoctoral Fellowship 1F32 HI 05108-01 (CVA)).

Title: Effect of Cold Acclimation on Oxygen Consumption in Muscle*

Authors: Barbara Grubb and G.E. Folk, Jr.

Journal: Journal of Comparative Physiology

Status: Submitted; twenty-three manuscript pages.

Coordination with Program: The previous page described a preliminary report of some of the information in this paper. We have singled out a description of the equipment and the data on the rats alone for initial publication. This manuscript has been sent to the editor of the Journal of Comparative Physiology, Dr. Ladd Prosser. The program now calls for continuing the same technique with more lemmings.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

Effect of Cold Acclimation on Norepinephrine
Stimulated $\dot{V}O_2$ in Muscle

The response to cold exposure in many mammals is characterized by two stages. During the acute stage, the animal increases its heat production by muscular contractions, i.e. by shivering. As cold exposure continues, shivering subsides yet the metabolic rate and thus heat production continue at a significantly elevated rate (Hart et al., 1956; Heroux et al., 1956; Mejsnar and Jansky, 1971b). There have been numerous theories proposed regarding both the site and mechanisms involved in this non-shivering thermogenic stage of cold acclimation. Evidence suggesting that norepinephrine mediates non-shivering thermogenesis (NST) in the cold acclimated animal is extensive (Bartunkova and Jansky, 1971; Hissa and Hirsimaki, 1971; Jansky et al., 1967; Leduc, 1961); however, the mechanisms of action remain unclear.

Animals with brown adipose tissue seem to have the greatest capacity for NST, yet it has been estimated that this tissue is responsible for a maximum of 6% of the extra heat produced in the cold (Jansky, 1971). Others have attributed 8.2% of the total heat production in the cold to brown adipose tissue (Imai et al., 1968). Thus, there must be large contributions of heat from tissue other than brown adipose tissue.

The role of the kidney, liver, and intestines has been examined and the combined heat production from these organs

Summary. The purpose of this study was to determine the effect of norepinephrine (NE) on oxygen consumption ($\dot{V}O_2$) of perfused (constant flow) muscle in cold acclimated (CA) and control rats. Infusion of NE for a five minute period caused an increase in $\dot{V}O_2$ of similar magnitude in both groups. Infusion of NE for 30 minutes resulted in an elevated steady state $\dot{V}O_2$ in the cold acclimated group, while the control group showed only an initial increase in $\dot{V}O_2$ followed by a continual decline during the remainder of the 30 minute infusion period. These results suggest that when rats are challenged by cold exposure, the magnitude of the initial muscle response to NE by control and acclimated rats is the same, but a useful sustained higher muscle oxygen consumption is found only in the cold acclimated animals.

Title: Daily Rhythm of Body Temperature of Wolverines (Gulo
luscus luscus)

Authors: G. Edgar Folk, Mary A. Folk, and Derek Craighead

Journal: The American Zoologist

Status: In Press; one page

Coordination with Program: This is an experiment of very long standing. We have been trying unsuccessfully for several years to develop a reasonably long-range temperature capsule. We finally tried one with the first two wolverines, and although a good experiment was completed, eventually because the animals were frightened and fought being put into Tim's respirometer, they broke these two transmitters. Fortunately no harm was done to the animals. The next two transmitters were different and stronger, and another successful experiment was accomplished. We plan and intend no more radio telemetry on any arctic mammals except lemmings. Nevertheless, we had already made body temperature observations at four times of the day on a series of weasels, and it is of the utmost interest to compare animals weighing 30 g to their nearest relatives which weigh 20 kg. This paper will be written this spring. A preliminary report is presented.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

DAILY RHYTHM OF BODY TEMPERATURE OF WOLVERINES
(*Gulo luscus luscus*)

by G. Edgar Folk, Jr., Mary A. Folk, and Derek Craighead
Naval Arctic Research Laboratory and The University of Iowa,
Iowa City, Iowa

The wide variation in size of species within the weasel family (from 30 grams to 20 kilograms) makes a comparison of their body temperatures more interesting. The body temperatures of four wolverines (maintained outdoors at Barrow, two in summer and two in winter), were recorded by Barrows radio-capsules implanted ventrally in the abdominal cavity. The resting body temperature of the wolverine was shown to change from a low to a higher setting approximately every 12 hours. Superimposed on these settings are body temperature increases due to activity. For example, the resting T_b of the two summer wolverines (designated as "Ne" and "M", both weighing 13.4 kg) was as follows (N=7): for Ne, low point each day was usually noon with a mean T_b of $38.27^{\circ}\text{C} \pm 0.24\text{SE}$; for M, low point usually 6 AM with a mean T_b of $37.99 \pm 0.11\text{SE}$. The peak of T_b and of activity for Ne was usually 6 AM with $38.56 \pm 0.39\text{SE}$; for M, usually midnight, with $38.22 \pm 0.13\text{SE}$. (Supported by The Arctic Institute of North America with approval and financial support of the Office of Naval Research under contract # N00014-75-C-0635 (Subcontract ONR-455).

Reference: American Zoologist 16: (in press) 1976.

Title: Use of Implants in Muskrat to Study Heart Rate and
Body Temperature*

Author: Keith E. Magoon

Journal: Master's Thesis

Status: Manuscript form; eighty-six pages, illustrated with
Kodachrome photographs.

Coordination with Program: This experiment developed with our Arctic program because Keith Magoon spent many months in my laboratory making the transmitters for use at NARL. The three EKG transmitters used in the arctic fox and in two wolves in the summer of 1975, were made by him. They proved to be greatly successful. While he was in my laboratory making these transmitters, he asked to do a Master's thesis during half time. Our plans were laid to have the thesis consist of these transmitters inserted into seals maintained at NARL. Over a two-year period, firstly, there was a delay in getting the permit for this, and secondly, the Utilidor broke down. For two reasons it seemed reasonable for him to do the thesis work on muskrats. In the first place, this is an aquatic animal and we were able to learn considerable information about the transmission from this transmitter through water to an antenna. This problem of course would come up with seals. Secondly, the major study on the seals would be that of diving bradycardia and of course we would obtain information on this from the muskrats. Thirdly, the muskrat can be classified as an Arctic animal since it is found on the edge of the Brooks Range on the Arctic Slope. This thesis is available for any interested individuals. There is a wealth of information in it, but unfortunately, Magoon I am sure will be very slow to contribute to the writeup and publication of this important material.

Credit: * Supported in part by The Arctic Institute of North America under contractual arrangements with the Office of Naval Research. We are also indebted to the director and staff of the Naval Arctic Research Laboratory for support and assistance throughout this investigation.

This study on muskrat, Ondatra zibethica, has revealed the following:

1. Body temperatures correlate with activity of the muskrat; lowest temperatures during sleep and rest and highest temperatures during activity.
2. Body temperature range of 99.2°F.-102.7°F. (37.3°C.-39.3°C.) is indicated.
3. A correlating rise in body temperature occurs when the animal acquires an infection.
4. Heart rates correlate with activity; lowest heart rates during rest and sleep and highest heart rates during activity.
5. A heart rate range of 64 to 250 plus beats per minute is indicated.
6. The effects of a severe injury were observed to increase heart rate and decrease the time spent in sleep, over a period of time.
7. Activity occurs during day and night, with a high per cent of a 24 hour period spent resting and sleeping.
8. Halothane was found to be a satisfactory anesthetic for the muskrat when dosage was kept under control.

ACKNOWLEDGEMENTS

The author is extremely grateful to the following persons:

Dr. G. Edgar Folk, Jr. for his most generous assistance and guidance throughout the investigation and preparation of this thesis and the funds he provided to help finish this thesis.

Dr. Lawrence E. Evans for his assistance with anesthetics during surgery and his advice throughout the investigation and preparation of this thesis.

John Copping for his guidance through the electronic problems of the EKG transmitters.

Dr. William D. Paul for his encouragement and guidance in preparation of this thesis.

Dr. Samuel M. McGinnis for his advice on temperature transmitters.

Dr. John H. Greve for analysis of the unique glandular tissue found in the stomach of the muskrat.

The author's parents (Virgil and Mary Magoon) for permitting the experiment to be conducted in their home.

The thesis committee (Dr. Alexander, Dr. Hall and Dr. Coppenger) for their guidance in the preparation of this thesis.

Mrs. Maxine Bogue for her help in preparing and typing this thesis.

ANNUAL REPORT - 1975

EXPENDITURES

Senior Investigator: G. Edgar Folk, Jr.

Part A: <u>Integrated Marine Mammal Plan</u>	\$ 756
Conferences with Elsner, Burns,	
Transportation, Meals, Phone	\$ 50
Obtaining Seal Permit, Secretary, Fee,	
Research Newsletter of Marine Mammal	
Conservation	75
Subscription to Marine Mammal News	35
Travel to NARL, G.E. Folk (March)	596
Facilities at NARL were not ready for seals in 1975	
Part B: <u>Body Temperature of the Wolverines</u>	2163
Transmitters (parts) Magoon	200
" (2) Barrows	600
Antenna wire and shipping	30
Travel to NARL, G.E. Folk (Nov.)	655
" " Mary Folk (Dec.)	678
Part C: <u>Rank Order, Breeding, Nutrition of Behavior Pack of Wolves</u>	2506
Travel to NARL, Mary Folk (March)	596
" " Katie Persons	600
Salary, " " July - Nov.	1090
Film and developing	20
4 Transmitters (glass & stainless steel)	200
Part D: <u>Fat Metabolism, Composition of Depot Fat Over Seasons</u> . . .	2022
Travel to NARL, Keith Magoon	643
Salary, " " May - Sept.	1040
Chemicals	190
Bio-mailers, insulated (12)	97
Instruments for fine dissection	52
Part E: <u>Cold Acclimatization of Lemmings: Isolated Muscles and</u>	
<u>Mechanism of Thermogenesis</u>840
Responsible Scientist: Dr. Barbara Grubb	
Travel to Durham, B. Grubb	210
Katie Persons (see above)	
Illustrations and photography	140
Surgical Supplies	30
Biochemical Supplies and Gas	90
Liquid nitrogen	90
Control white rats	148
European lemmings, 6 @ \$7	42
Lemmings, Barrow, shipping and returning box	90

EXPENDITURES 1975 (continued)

Part F: <u>Cold Acclimatization of Lemmings: Mechanism of Increased</u>		
<u>Tolerance to -40° Ambient</u>		\$5432
Travel to NARL, G.E. Folk (Dec.)	\$ 678	
" " Bill Barrows (July)	540	
" " G.E. Folk (July)	678	
" " Peter Ringens (July)	847	
" " Dr. O. Héroux (July)	852	
" " Edward Koo (July, Aug.)		
(Paid own way to Fairbanks)	660	
Salary, Edward Koo	300	
Shipping equipment, Ottawa, Barrow	126	
" lemmings to Iowa & to Ottawa	60	
Respirometer	245	
Cool-Pax	30	
2 Aquarium pumps	16	
Transmitters, Barrows (temperature)	400	
Part G: <u>Extension of Lemming Program: Circadian Rhythms in</u>		
<u>Continuous Light, True Microclimate of Freelifving</u>		
<u>Lemmings, Deacclimatization After Cold Exposure</u>		738
Travel to NARL, Mary Folk (July)	678	
Repair radiometers	40	
" stopwatch	20	
Part H: <u>Kidney Function of Polar Bears</u>		200
Anna Larson, Arctic Medical Research Laboratory,		
Chemistry		
Transportation and shipping samples	200	
Part I: <u>Hibernation Factor</u>		670
Responsible Scientist: Martin Steiner		
Travel to Iowa, Wilma Spurrier	80	
Animal cages	120	
50 Hamsters	150	
Animal care (marmots and hamsters)	200	
2 temperature probes	100	
Biochemicals	20	
Part J: <u>Miscellaneous</u>		5673
1. Dan Nuno (hourly pay)	475	
2. Karen Hagelstein (hourly pay)	180	
3. Louise Janes (hourly pay)	1943	
4. 14 trips/diems (28 x \$25)	700	
5. Reprints, Joel, lemming paper	140	
6. Reprints & page charges, cardiovascular paper	150	
7. Biometeorology Meeting	289	
8. Telephone	442	
9. Office Supplies	97	
10. Xerox	213	
11. Postage	346	
12. Recording paper	300	
13. Reprints on order, kidney paper	398	

\$21,000

Item 15. Explanation of Expenditures

The major items and experimental blocks have been explained in the Introduction of this report. During the summer of 1975, most of the "team-energy" went toward a cooperative experiment with the National Research Council of Canada. The Canadians had done earlier experiments on white rats in which they found a broad range of response of rats when exposed to -40° . This was in spite of the fact that the rats were carefully selected and standardized. They have been trying to study the early failures and the very tolerant rats in order to explain what physiological contributions make some of these animals more resistant. The Canadians were very eager to make a comparison with their white rats and some arctic rodents. For some time, we have been trying similar experiments. Therefore, the National Research Council sent Dr. Oliver Héroux to work with us in the summer (see Expenditures, Part F). After this work in the summer, during the fall and winter, much of our time was spent on Part E, the Isolated Muscle experiment (see illustrations at end of report). Specific items will now be explained as follows:

Part B, Transmitters, \$600. These transmitters were made in California by William Barrows. They consisted of four body temperature radio-capsules with a long life that could be read at a distance of 100 yards. We were very fortunate to have these transmitters developed, since they are unique. Many other industries associated with making radio-capsules would have charged \$1,000 each. Two of these are still functioning in two of the wolverines.

Part C, Travel for Katie Persons, \$600. This item includes not only her round trip fare to NARL, but one trip to Fairbanks which we sponsored because we wished her to obtain training at the Institute of Arctic Biology in taking out pineal glands of rodents. This trip was arranged at the end of five months at Point Barrow and provided well-earned and well-justified refreshment for her as well as training. Her salary consisted of \$218 per month for five months, from July through November.

Part C, Transmitters, \$200. These excellent transmitters were made by Keith Magoon and data were obtained with them from not only foxes and wolves, but also from a diving mammal. Because NARL was unable to maintain seals, we did a preliminary experiment on muskrats, which resulted in a Masters thesis. Note that muskrats occur on the Arctic Slope.

Part D, Keith Magoon, Salary. For five months, Keith was paid \$210 a month. During this time he made four excellent radio-capsules which performed without failure. His biggest contribution, however, was repairing with considerable difficulty, the freeze-dry equipment at the Naval Arctic Research Laboratory. This had been left in broken and extremely bad condition by the last investigator. Essentially, Magoon's entire summer was spent trying to repair, nurse along, and maintain this equipment. He not only used it for fat samples from two seals obtained from the Arctic ice pack (it was necessary to take dry ice and nitrogen out to the pack), but he also did freeze-drying of many hundreds of samples from lemmings.

Item 15, Explanation of Expenditures (continued)

Part E, Dr. Grubb, Travel. This trip was to Durham, N.C. to visit the laboratory of Professor Schmidt-Nielsen at Duke University. Dr. Grubb had been having difficulty with the perfusion (standardized) solution which has to be used through vessels maintaining the hind limb of animal preparations. She obtained information from the laboratory where she visited which resulted in excellent preparations back here at The University of Iowa. When the lemming and rat preparations have been completed at this university, Dr. Grubb will carry out the same technique in the laboratory of Professor Schmidt-Nielsen. This will allow coordination between The University of Iowa and Duke University which will be mutually beneficial.

Part E, Photography. Photography for the year 1975 represents a very high figure, only part of which is charged to this grant. One reason was that Dr. Denner requested samples in the form of slides of each of the projects done at NARL. It was expensive having these slides copied and sent to the Laboratory, and part of these charges must be included in 1976. Note, however, the illustrations of the equipment at the end of this report describing the method of perfusing and maintaining isolated muscles.

Part F, Travel for Bill Barrows. This engineer brought the wolverine transmitters and the lemming transmitters. We were very familiar with the small lemming transmitters and I did not have any doubts about using them. The large wolverine transmitters were a new development, however, and from past experience I knew that the only procedure which would be safe would be to have the engineer on the spot when these were implanted in the wolverines. We did bring in William Barrows and it was most fortunate that he was here since one of the transmitters failed before it was even put in the wolverine. He fixed it on the spot and all went well from that point on. Furthermore, he demonstrated the correct way to calibrate these transmitters. Because many other industries would have charged us \$1,000 for each transmitter, the cost of his airplane transportation from California is much more than justified.

Part F, Ringens, Travel. This biochemist had been with us the summer before. We needed him very badly but his airplane fare round trip was more expensive than our own (\$847). We told him we would not be able to have him with us unless he would serve without salary. He very willingly did this and a nice paper described within this report is the result of his work.

Part F, Edward Koo. This senior at Amherst College received many complimentary letters from the faculty of that school. We needed extra help but did not feel we could take a chance with this student who was determined to work in the Arctic area. We insisted that he pay his own way to Point Barrow and would serve without salary. As it turned out, he was a brilliant individual with a flair for repairing complicated equipment. The metabolic self-recording equipment sent from Canada by the National Research Council, had many bugs

Item 15, Explanation of Expenditures (continued)

in it. This student did a fine job of removing these difficulties and making the equipment work. In addition to that, he turned out to be a first-rate statistician. By the end of the summer, he was the leader in each daily complex experiment. A difficulty arose in sending our samples from lemmings through Canadian customs, so that they could be analyzed at the National Research Council in Ottawa. We finally paid the way home of Eddie Koo from NARL carrying many hundreds of samples through customs to the NRC of Canada from which he continued on to Amherst. In addition to this, his services had been so outstanding, we paid him \$150 for each of the two months he was with us.

Part F, Transmitters, Temperature, William Barrows. These transmitters have been discussed above; they are long life temperature capsules weighing about 4 grams, that we have used before. We obtained 5 of them from the Barrows Co. at \$80 each. This is a remarkably small charge for this reliable temperature transmitter.

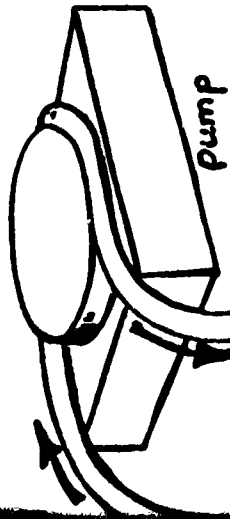
Part I, Travel to Iowa, Spurrier. This took place because of a very complex experiment. We had collected hibernating blood from marmots and wanted to dialyze one-half of it here at Iowa. The other half was to be dialyzed by the expert in this field, Wilma Spurrier, in the laboratory of Professor Albert Dawe. In addition to this, she was to teach us to inject this material into our test animals (ground squirrels) by means of the saphenous vein. This is a very difficult procedure. She came out to The University of Iowa with the dialyzed material and our mission was accomplished.

Part J, Item 1. This pay was for animal maintenance.
" " 2. This pay was for measuring the pineal glands of lemmings.
" " 3. This pay was for acting as Laboratory Manager, typing reports and publications, supervising animal care and actually doing animal care.
" " 4. These expenses were because each individual who travelled through Fairbanks on the way to NARL had to spend one night on the way there, and usually one night on the way upon return. You will note, however, that on numerous occasions some of us were able to stay at the house of a friend to avoid the expense of a hotel. Per diems are charged for approximately 1/2 of the trips.
" " 7. This expense was to the International Congress of Biometeorology and for a conference with Dr. Arthur Callahan. The principal investigator had arranged a symposium which included material on arctic pineal glands (see Item 7 in Table of Contents).
" " 12. We have been very successfully recording the EKG by means of our radio-capsules. Although this has not been mentioned to date, steady progress is being made on the analysis of the EKG of at least six arctic mammals. This analysis continued all through 1975. One half of this recording paper as mentioned is available for the work in 1976.

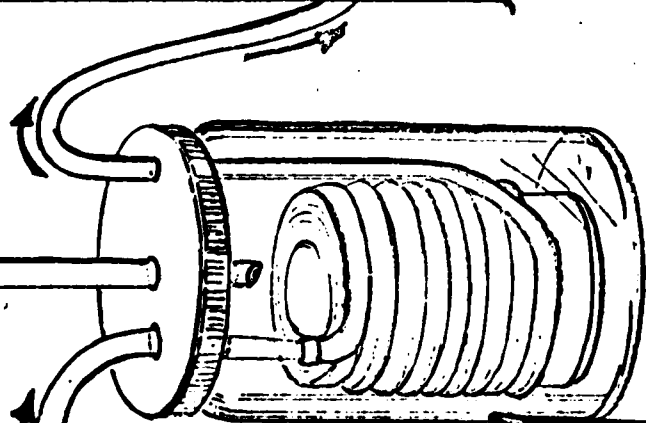
Item 16. Illustrations

The following three illustrations explain the apparatus and technique for perfusing isolated muscles so that norepinephrine can be administered in experiments to explain the mechanism of thermogenesis.

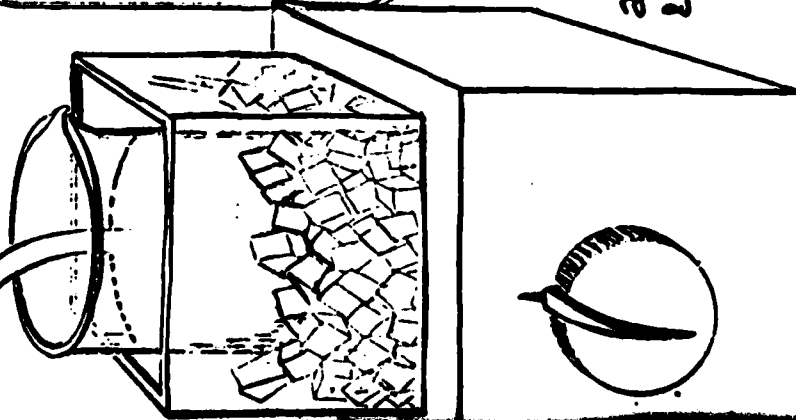
O₂-CO₂



pump

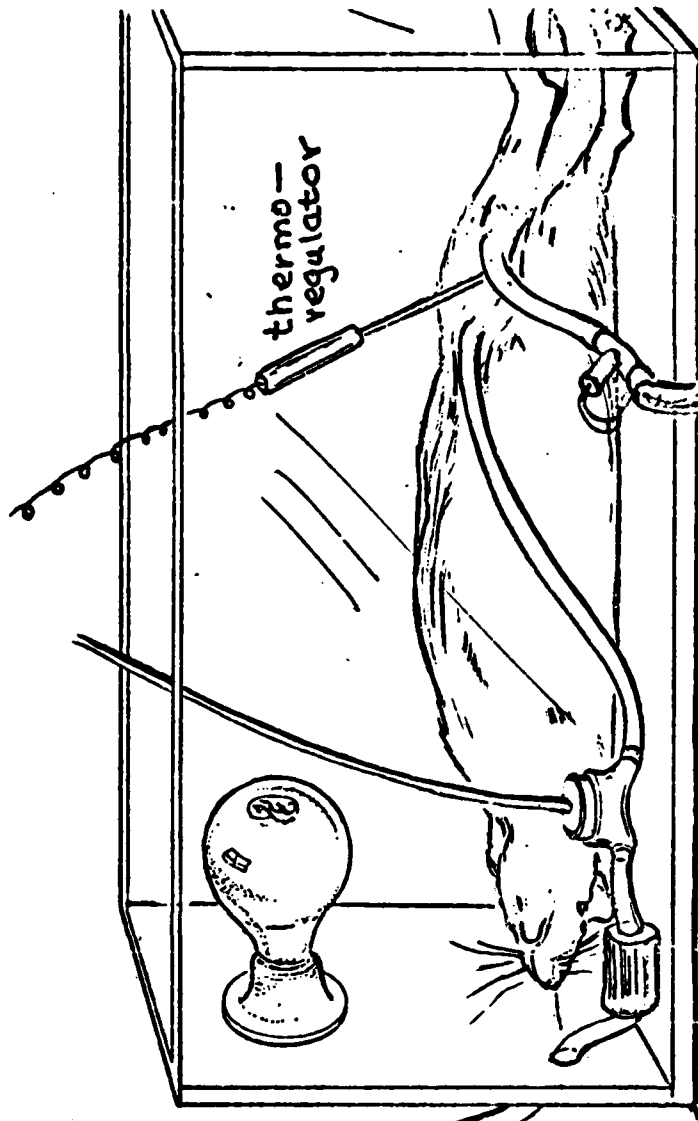


oxygenator



arterial reservoir
& magnetic stirrer

to transducer

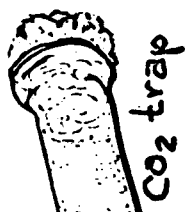


thermo-
regulator

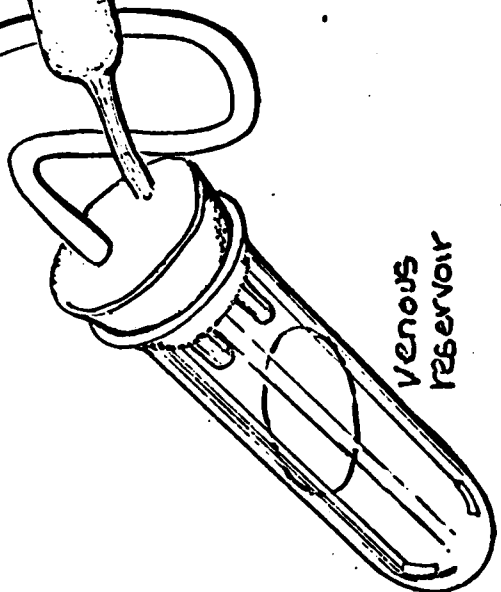
O₂ electrode

bubble
trap

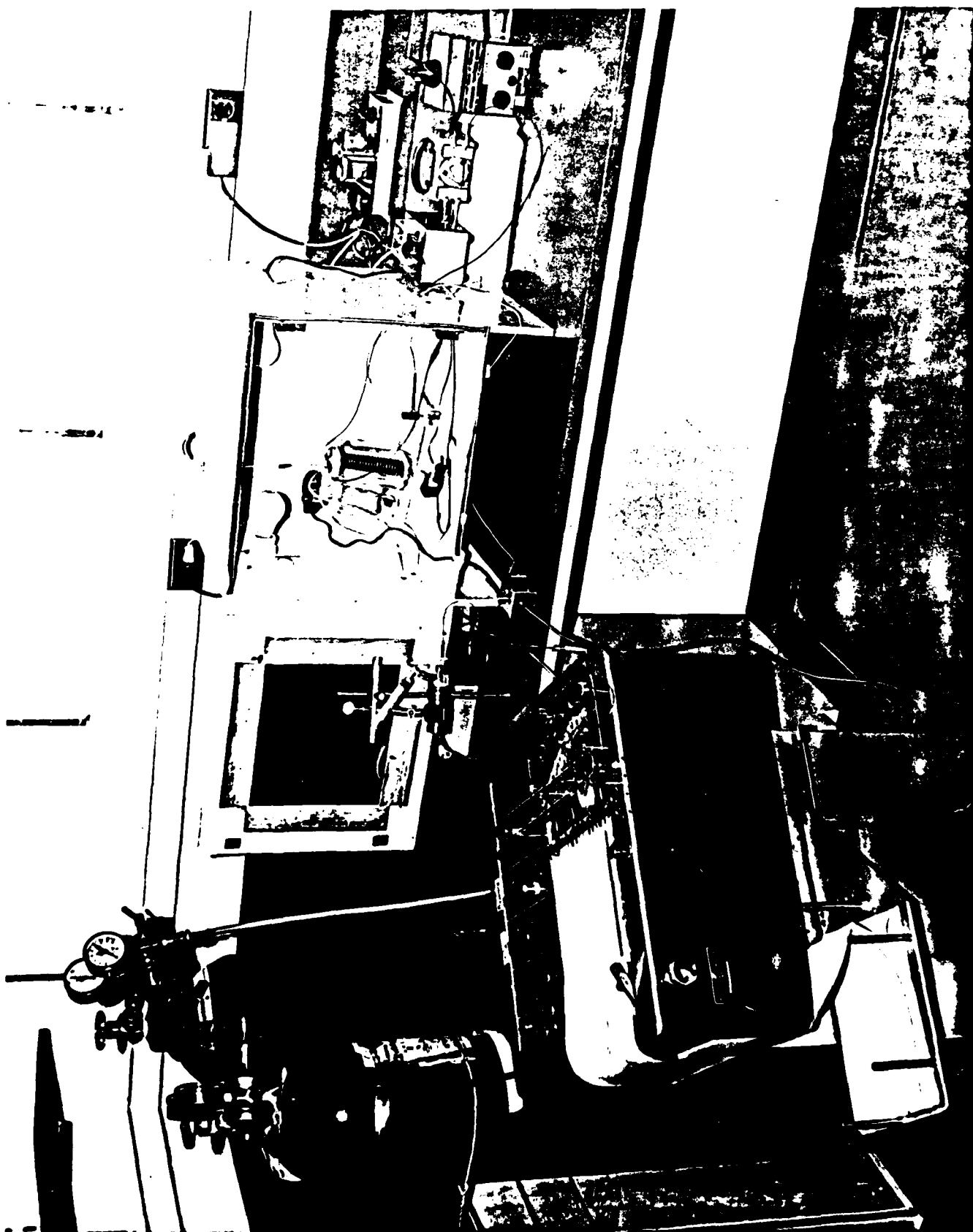
filter

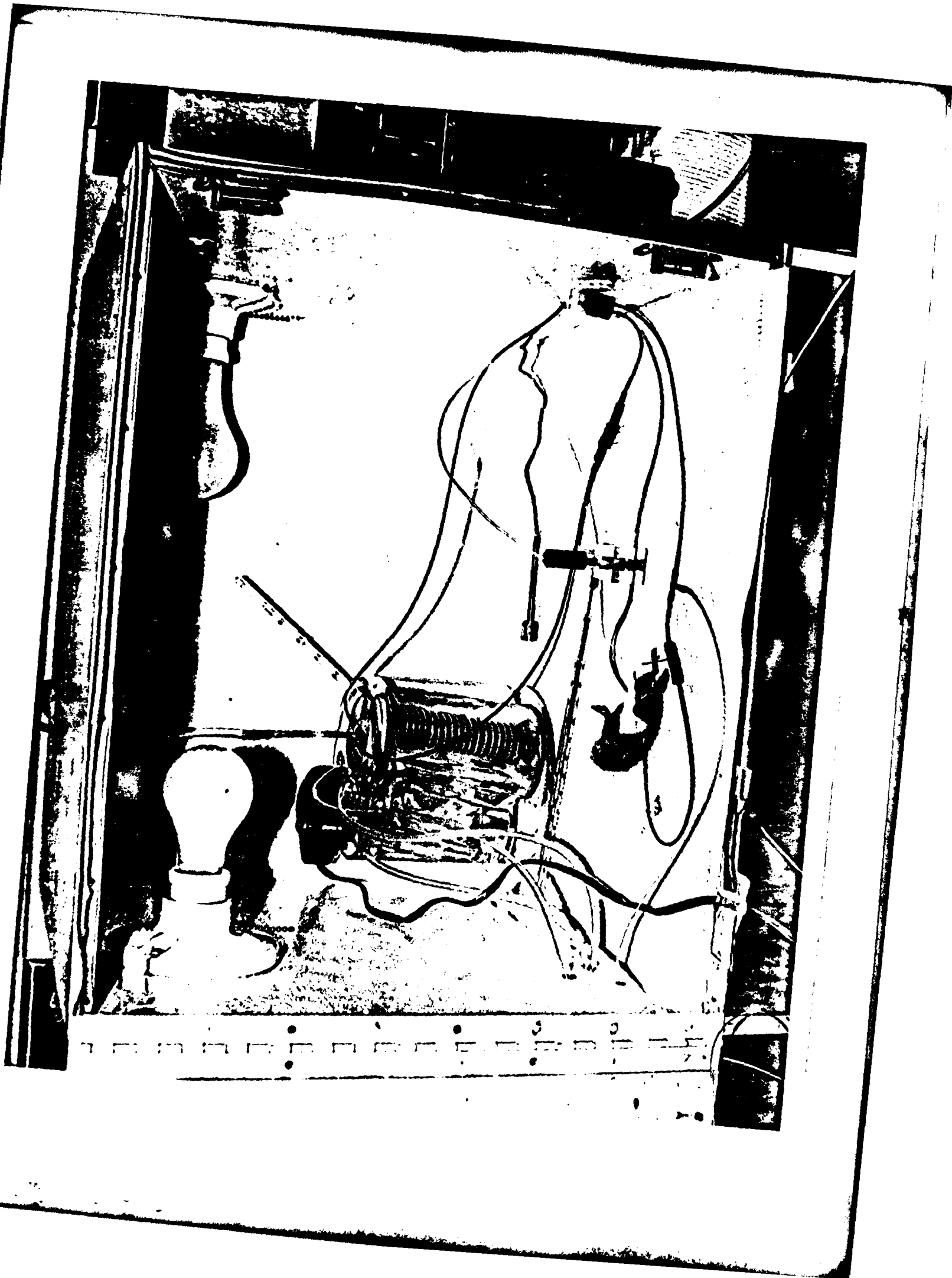


CO₂ trap



venous
reservoir





6F